USE OF BISPHOSPHONIC ACID DERIVATIVES FOR THE TREATMENT OF CALCIUM PYROPHOSPHATE DEPOSITION DISEASE AND DENTAL TREATMENT

Field of The Invention

The present invention relates to the use of bisphosphonic acid derivatives for the preparation of a medicament for the treatment of calcium pyrophosphate deposition disease (CPDD) primarily pseudogout and chondrocalcinosis, in a mammal. More specifically, the present invention relates to the use of bisphosphonic acid derivatives, said derivatives being especially adapted to be administered to subjects suffering from CPDD. The invention also relates to the use of bisphosphonic acid derivatives for the prevention or treatment of caries, such as secondary caries.

Background of The Invention

The term "calcium pyrophosphate deposition disease" (CPDD) is used to describe the clinical syndrome of acute gout-like arthritis associated with the presence of calcium pyrophosphate crystals in the synovial fluid of a mammal. Other terms used to define the clinical syndrome of acute gout-like arthritis are: "Calcium pyrophosphate dihydrate crystal deposition disease" (CPPD Disease), "pyrophosphate gout" or "pseudogout".

CPDD is seldom seen in patients below age 50; the overall incidence of this disease occurs in the later years of life. The most common association of calcium pyrophosphate crystal deposition is the formation of these crystals in the joints of aged patients (Kenneth P.H. Pritzker, "Crystal-associated Arthropathies: What's New on Old Joints", J. American Geriatrics Society, 28 (1980) 439-445).

The calcium pyrophosphate crystal deposits are topologically confined to the hyaline cartilage, the fibrocartilage in the meniscus of the knee, the annulus fibrosus of the intervertebral disc, the synovial fluid, or the synovium and tendon insertions (Kenneth P.H. Pritzker et al., "Crystal-associated Arthropathies: What's New on Old Joints", J. American Geriatrics Society, 28 (1980) 439-445).

The calcium pyrophosphate crystal deposits are also frequently known to form or precipitate in the articular cartilage, particularly in elderly people (Ryan, L.M. et al., "Calcium pyrophosphate crystal deposition disease, pseudogout and articular chondrocalcinosis", In: Arthritis and Allied Conditions, A Textbook in Rheumatology, 13th edition, Vol, II; Ed. W.J. Kooperman, 1997, pp. 2103-2125).

The calcium pyrophosphate crystal deposits are known to especially form in the synovial fluid, particularly in elderly people.

The formation of calcium pyrophosphate crystal deposits is believed to be caused by an increase in the concentration of pyrophosphate, PP, caused by the changes in the PP metabolism of chondrocytes (Ryan et al., "Understanding inorganic pyrophosphate metabolism: toward prevention of calcium pyrophosphate dihydrate crystal deposition, Annals of the Rheumatic Diseases 54 (1995) 939-941).

The disodium salt of ethane-1-hydroxy-1,1-bisphosphonic acid, abbreviated EHDP, and a series of similar bisphosphonates are used in the treatment of osteoporosis and for the prevention of bone fractures in connection to various cancer diseases. Such compounds are known under various trade names, e.g. "Didronel" or "Aredia" or "Fosamax".

EP 0924293 discloses a fabric care composition including the compound hydroxy-ethane-1,1-diphosphonic acid (HEDP/EDHP) and its use in order to inhibit the formation of inorganic microcrystals.

US 3 683 080 discloses a composition which may include effective amounts of polyphosphonates, such as ethane-1-hydroxy-1,1-bisphosphonic acid (EHDP), for the inhibition of anomalous deposition and mobilisation of calcium phosphates in animal tissue.

EP 563096 discloses anti-inflammatory compositions comprising salicylic acid-, phenylacetic acid-, anthranilic acid- based inflammation inhibitors and an amount of an organic phosphonic acid or one of its salts or esters, said compositions being suitable for treating rheumatoid arthritis, bone infections and bone degradation.

US 4 812 304 discloses a method for treating or preventing osteoporosis in humans, the method amongst others comprising a bone resorption period during which ethane-1-hydroxy-1,1-diphosphonic acid (EHDP) is administered.

US 5 882 656 discloses pharmaceutical compositions of bisphosphonic acids for the treatment of disturbances involving the calcium or phosphate metabolism. Suitable organophosphonate compounds include 4-amino-1-hydroxybutylidene-1,1-bisphosphonic acid.

US 6 221 861 discloses a method for the treatment of an animal with pyrophosphate gout comprising administering an effective amount of calcium antagonists, such as phenylalkylamines, dihydropyridines or benzothiazepines.

JP 10017493 discloses an external anti-inflammatory or antiallergic skin composition containing at least one calcium ion blocking agent consisting of hydroxyethanediphosphonic acid (EHDP).

Summary of The Invention

A first object of the invention is directed to the use of a bisphosphonic acid derivative for the preparation of a medicament for the treatment of calcium pyrophosphate deposition disease (CPDD) in a mammal.

A related aspect of the invention relates to a composition for the delaying the deposition of calcium pyrophosphate depots in hyaline cartilage, the fibrocartilage in the meniscus of the knee, the annulus fibrosus of the intervertebral disc, the synovial fluid, or the synovium and tendon insertions, comprising a 1,1-bisphosphonic acid derivative.

A further aspect of the invention relates to the use of etidronate, pamidronate, alendronate, tiludronate, risedronate, zoledronic acid, clodronic acid or ibandronic acid for the preparation of a medicament for the treatment of calcium pyrophosphate deposition disease (CPDD) in a mammal.

A second object of the invention relates the use of a bisphosphonic acid derivative for the manufacture of a medicament for the prevention or treatment of secondary caries or for the treatment of persons suffering form primary caries.

An interesting aspect of the invention related to the use of etidronate, pamidronate, alendronate, tiludronate, risedronate, zoledronic acid, clodronic acid or ibandronic acid for the preparation of a medicament for the prevention or treatment of secondary caries or for the treatment of a mammal suffering from primary caries.

Brief Description of The Invention

In spite of the substantial body of literature relating to the component of interest within the present invention, such as ethane-1-hydroxy-1,1-bisphosphonic acid (EHDP), the use of said compound and its acid derivatives for the treatment of Calcium Pyrophosphate Deposition Disease (CPDD) or pseudogout does not appear to have been appreciated heretofore.

According to the present invention, bisphosphonate acid derivatives, such as alkyl-1,1-bisphosphonic acids (EHDP), have the ability to severely inhibit the rate of growth of various forms of calcium pyrophosphate crystals when present in medically relevant concentrations. A particular embodiment of the present invention, as will be shown in the following, is the ability to severely retard the spontaneous precipitation of various forms of

calcium pyrophosphate crystals from solutions supersaturated with respect to calcium pyrophosphate. Rates of dissolution are however unchanged.

According to the present invention, bisphosphonates can be used in yet another context, namely for the manufacture of a medicament for the prevention or treatment of secondary caries, i.e. caries that forms at the interface between the natural dental material (enamel, dentine, cementum and root material) and the filling material. Experiments indicate that the use of bisphosphonates in this context is able to inhibit or reduce the development of secondary caries in an animal, preferably a human. It is believed that EHDP acts as a chelating agent and thereby reduces or inhibits the dissolution of tooth minerals beneath or in the vicinity of the "primary" caries under repair. Hence, use of a bisphosphonate prior to any medical operation or procedure on a tooth subject to "primary caries" will have the advantage of providing a stable tooth in which substantially no secondary caries will be susceptible to form. When EHDP is applied to a tooth subject to "primary caries", it will result in, amongst others, an unchanged adhesion between the natural dental material (enamel, dentine, cementum and root material) and the filling material, as compared with a similar tooth that has not been subjected accordingly with EHDP.

Description of The Invention

The present invention provides use of an effective amount of a bisphosphonic acid derivative, for the preparation of a medicament for the treatment of calcium pyrophosphate deposition disease (CPDD) in an animal. The animal is preferentially a human. As used herein, CPDD includes pseudogout, chondrocalcinosis, and any other disease caused by the deposition of calcium pyrophosphate crystals in the body. The invention relates, in one aspect, to a method of treating or preventing calcium pyrophosphate deposition disease (CPDD), such as pseudogout and chondrocalcinosis in a mammal.

The term "bisphosphonic acid derivative" when referred to herein are intended to mean bisphosphonic acid derivatives, salts thereof, esters thereof or hydrates thereof.

The bisphosphonic acid derivatives of the present invention are discussed more fully hereinafter. The term "bisphosphonic acid derivative" is intended to mean a compound having two phosphonic acid groups bound to the same carbon atom, namely 1,1-bisphosphonic acid derivative (geminal bisphosphonic acid derivatives), as well as salts or

hydrates thereof. Thus, the invention relates to the use of a compound comprising a -$C(PO_3H_2)_2$- moiety, or a pharmaceutically acceptable salt or hydrate of said compound.

Bisphosphonic acid derivatives useful herein are of the formula

$$R^1\text{-}C(PO_3H_2)_2\text{-}R^2 \qquad (I)$$

wherein R^1 and R^2 are selected from selected from any array of substituents, preferably wherein R^1 and R^2 comprise an electronegative group.

Suitably, R^1 and R^2 may be independently selected from hydrogen, halogen, COOH, optionally substituted $C_{1\text{-}12}$-alkyl, optionally substituted aryl, optionally substituted $C_{3\text{-}9}$-cycloalkyl, optionally substituted heterocyclyl, optionally substituted heteroaryl, optionally substituted $C_{1\text{-}12}$-alkyl-aryl, optionally substituted $C_{1\text{-}12}$-alkyl-$C_{3\text{-}9}$-cycloalkyl, optionally substituted $C_{1\text{-}12}$-alkyl-heteroaryl, heteroaryl, heterocyclyl, optionally substituted $C_{1\text{-}12}$-alkyl-heterocyclyl, amino, optionally substituted $C_{1\text{-}12}$-alkyl-amino, optionally substituted amino-$C_{1\text{-}12}$-alkyl, optionally substituted amino-$C_{3\text{-}9}$-cycloalkyl, optionally substituted $C_{1\text{-}12}$-alkyl-halide, optionally substituted $C_{1\text{-}12}$-alkyl-OH, optionally substituted $C_{1\text{-}12}$-alkyl-SH, alkoxy, optionally substituted $C_{1\text{-}12}$-alkyl-O-alkyl, $C_{1\text{-}12}$-alkyl-S-alkyl, optionally substituted $C_{1\text{-}12}$-alkyl-COOH, and optionally substituted $C_{1\text{-}12}$-alkyl-PO_3H_2.

The term "optionally substituted $C_{1\text{-}12}$-alkyl" in itself or when used as a moiety within a group, is intended to mean an optionally substituted alkyl chain of 1-12 carbons in length. Suitably the optionally substituted $C_{1\text{-}12}$-alkyl" is a $C_{1\text{-}6}$-alkyl having 1-6 carbon atoms.

A substituted $C_{1\text{-}12}$-alkyl may have a substituent any position along the alkyl chain. A substituent may be of any array known to the person skilled in the art, such as a halogen, COOH, optionally substituted $C_{1\text{-}12}$-alkyl, optionally substituted aryl, optionally substituted $C_{3\text{-}9}$-cycloalkyl, optionally substituted heterocyclyl, optionally substituted heteroaryl, optionally substituted $C_{1\text{-}12}$-alkyl-aryl, optionally substituted $C_{1\text{-}12}$-alkyl-$C_{3\text{-}9}$-cycloalkyl, optionally substituted $C_{1\text{-}12}$-alkyl-heteroaryl, heteroaryl, heterocyclyl, optionally substituted $C_{1\text{-}12}$-alkyl-heterocyclyl, amino, optionally substituted $C_{1\text{-}12}$-alkyl-amino, optionally substituted amino-$C_{1\text{-}12}$-alkyl, optionally substituted $C_{1\text{-}12}$-alkyl-halide, optionally substituted $C_{1\text{-}12}$-alkyl-OH, optionally substituted $C_{1\text{-}12}$-alkyl-SH, optionally substituted O-$C_{1\text{-}12}$-alkyl, optionally substituted $C_{1\text{-}12}$-alkyl-O-alkyl, $C_{1\text{-}12}$-alkyl-S-alkyl, optionally substituted $C_{1\text{-}12}$-alkyl-COOH, and optionally substituted $C_{1\text{-}12}$-alkyl-PO_3H_2.

An amine may be optionally substituted in any manner known to the person skilled in the art, such as with a COOH, optionally substituted $C_{1\text{-}12}$-alkyl, optionally substituted aryl, optionally substituted $C_{3\text{-}9}$-cycloalkyl, optionally substituted heterocyclyl, optionally

substituted heteroaryl, optionally substituted C_{1-12}-alkyl-aryl, optionally substituted C_{1-12}-alkyl-C_{3-9}-cycloalkyl, optionally substituted C_{1-12}-alkyl-heteroaryl, heteroaryl, heterocyclyl, optionally substituted C_{1-12}-alkyl-heterocyclyl, amino, optionally substituted C_{1-12}-alkyl-amino, optionally substituted amino-C_{1-12}-alkyl, optionally substituted C_{1-12}-alkyl-halide, optionally substituted C_{1-12}-alkyl-OH, optionally substituted C_{1-12}-alkyl-SH, optionally substituted O-C_{1-12}-alkyl, optionally substituted C_{1-12}-alkyl-O-alkyl, C_{1-12}-alkyl-S-alkyl, optionally substituted C_{1-12}-alkyl-COOH, and optionally substituted C_{1-12}-alkyl-PO_3H_2

The term "optionally substituted C_{1-12}-alkyl-amino " is intend to mean an optionally substituted alkyl chain of 1-12 carbons in length with an amino group within or at the end of said chain. The amino group may be a free amino or optionally substituted. The term "optionally substituted amino-C_{1-12}-alkyl" is intended to mean an amino group bound to the central carbon of the bisphosphonic acid moiety, said amino group further bound to an optionally substituted alkyl chain of 1-12 carbons in length.

The term "optionally substituted C_{1-12}-alkyl-C_{3-9}-cycloalkyl" is intended to mean an optionally substituted alkyl chain of 1-12 carbons in length with an optionally substituted C_{3-9}-cycloalkyl at the end. The other terms for substituents are to be understood in the same manner as defined for the term "optionally substituted C_{1-12}-alkyl-C_{3-9}-cycloalkyl".

The term "halogen" is intended to mean that R^1 and R^2 may be independently selected from Cl, Br, F and I.

Typically, R^1 may be selected from hydrogen, halogen, -COOH, optionally substituted C_{1-12}-alkyl, optionally substituted C_{3-9}-cycloalkyl, optionally substituted heterocyclyl, optionally substituted heteroaryl, optionally substituted C_{1-12}-alkyl-aryl, optionally substituted C_{1-12}-alkyl-heteroaryl, heteroaryl, heterocyclyl, optionally substituted C_{1-12}-alkylheterocyclyl, optionally substituted C_{1-12}-alkyl-C_{3-9}-cycloalkyl, amino, optionally substituted C_{1-12}-alkyl-amino, optionally substituted amino-C_{1-12}-alkyl, optionally substituted C_{1-12}-alkyl-halide, optionally substituted C_{1-12}-alkyl-OH, alkoxy, and optionally substituted C_{1-12}-alkyl-O-alkyl; and R^2 may be selected from hydrogen, halogen, -COOH, optionally substituted C_{1-12}-alkyl, optionally substituted C_{3-9}-cycloalkyl, optionally substituted heterocyclyl, optionally substituted heteroaryl, optionally substituted C_{1-12}-alkyl-aryl, optionally substituted C_{1-12}-alkyl-heteroaryl, heteroaryl, heterocyclyl, optionally substituted C_{1-12}-alkyl-heterocyclyl, optionally substituted C_{1-12}-alkyl-C_{3-9}-cycloalkyl, amino, optionally substituted C_{1-12}-alkyl-amino, optionally substituted amino-C_{1-12}-alkyl, optionally substituted C_{1-12}-alkyl-halide, optionally substituted C_{1-12}-alkyl-OH, alkoxy, optionally substituted C_{1-12}-alkyl-O-alkyl, optionally substituted C_{1-12}-alkyl-COOH, and optionally substituted C_{1-12}-alkyl-PO_3H_2.

In a further typical embodiment R^1 is selected from the group consisting of hydrogen, halogen, hydroxy and R^2 is selected from hydrogen, halogen, COOH, optionally substituted C_{1-12}-alkyl, optionally substituted aryl, optionally substituted C_{3-9}-cycloalkyl, optionally substituted heterocyclyl, optionally substituted heteroaryl, optionally substituted C_{1-12}-alkyl-aryl, optionally substituted C_{1-12}-alkyl-C_{3-9}-cycloalkyl, optionally substituted C_{1-12}-alkyl-heteroaryl, heteroaryl, heterocyclyl, optionally substituted C_{1-12}-alkyl-heterocyclyl, amino, optionally substituted C_{1-12}-alkyl-amino, optionally substituted amino-C_{1-12}-alkyl, optionally substituted C_{1-12}-alkyl-halide, optionally substituted C_{1-12}-alkyl-OH, optionally substituted C_{1-12}-alkyl-SH, alkoxy, optionally substituted C_{1-12}-alkyl-O-alkyl, C_{1-12}-alkyl-S-alkyl, optionally substituted C_{1-12}-alkyl-COOH, and optionally substituted C_{1-12}-alkyl-PO_3H_2.

In a suitable embodiment, R^1 is selected from hydrogen, halogen, -COOH, alkoxy, C_{1-12}-alkyl, optionally substituted C_{1-12}-alkyl-amino, optionally substituted amino-C_{1-12}-alkyl, optionally substituted C_{1-12}-alkyl-O-alkyl, optionally substituted C_{1-12}-alkyl-hydroxy, and optionally substituted C_{1-12}-alkyl-halide, and R^2 is selected from hydroxy, halogen, COOH, amino, optionally substituted C_{1-12}-alkyl-amino, amino-C_{3-9}-cycloalkyl, optionally substituted amino-C_{1-12}-alkyl, optionally substituted C_{1-12}-alkyl-O-alkyl, optionally substituted C_{1-12}-alkyl-hydroxy, optionally substituted C_{1-12}-alkyl-heterocyclyl, optionally substituted C_{1-12}-alkyl-halide, optionally substituted C_{1-12}-alkyl-COOH, and optionally substituted C_{1-12}-alkyl-PO_3H_2.

In a further suitable embodiment, R^1 is selected from the group consisting of halogen, hydroxy, amino, and R^2 is a halogen, such as a chloro group. In a further suitable embodiment one or both of R^1 and R^2 is a hydroxy group. In a further suitable embodiment one or both of R^1 and R^2 is an optionally substituted alkyl amino group. In a combination of suitable embodiments, one of R^1 and R^2 is hydroxy and the other and R^2 is a substituted amino alkyl group, such as in ibandronic acid (1-hydroxy-3[methyl(pentyl) aminopropylidene]1,1-bisphosphonic acid) or salts or hydrates thereof.

In a suitable embodiment, one or both of R^1 and R^2 is a C_{1-12}-alkyl-heterocyclyl such as an C_{1-12}-alkyl-pyrrolidene, such as a C_{1-6}-alkyl-pyrrolidene such as a propyl-pyrrolidene, such as [1-Hydroxy-3-(1-pyrrolidinyl)propylidene]bisphoshonate or salts or hydrates thereof.

In a further suitable embodiment, the bisphosphanate is (6-Amino-1-hydroxyhexylidene)bisphosphonate (neridronate) or salts or hydrates thereof.

In a further suitable embodiment, one or both of R^1 and R^2 is an optionally substituted alkyl amino group, such as an alkyl (dialkyl)amino group, such as the bisphosphonate is

[3-(Dimethylamino)-1-hydroxypropylidene]bisphosphonate (olpadronate) or salts or hydrates thereof.

In a further suitable embodiment, one or both of R^1 and R^2 is a C_{1-12}-alkyl-heteroaryl, such as [1-Hydroxy-2-imidazo-(1,2a)pyridin-3-ylethylidene]bisphosphonate (minodronate) or salts or hydrates thereof.

In a further suitable embodiment, one or both of R^1 and R^2 is an optionally substituted amino-C_{3-9}-cycloalkyl, such as [(Cycloheptylamino)-methylene]bisphosphonate, (incadronate) or salts or hydrates thereof.

In a still further suitable embodiment, R^1 is selected from hydrogen, halogen, amino, and C_{1-12}-alkyl, and C_{1-12}-alkylamino and R^2 is selected from hydrogen, halogen, hydroxy, amino, C_{1-12}-alkylamino, $-CH_2COOH$, $-CH_2PO_3H_2$ and - $CH_2CH_2PO_3H_2$.

In a still further suitable embodiment, R^1 is selected from hydrogen and C_{1-12}-alkyl, and R^2 is selected from hydroxy, amino, $-CH_2COOH$, $-CH_2PO_3H_2$ and - $CH_2CH_2PO_3H_2$.

In a suitable embodiment of the present invention, R^1 is selected from C_{1-12}-alkyl and R^2 is selected from hydroxy.

In a further suitable embodiment of the present invention, R^1 is selected methyl and R^2 is selected from hydroxy.

Alternatively stated, one aspect of the invention is directed to a composition for the delaying the deposition of calcium pyrophosphate depots in hyaline cartilage, the fibrocartilage in the meniscus of the knee, the annulus fibrosus of the intervertebral disc, the synovial fluid, or the synovium and tendon insertions, comprising a bisphosphonic acid derivative, as defined supra.

Alternatively stated, one aspect of the invention is directed to a composition for the treatment of pseudogout or chondrocalcinosis.

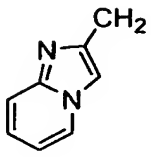
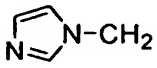
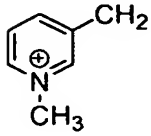
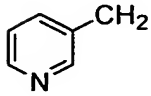
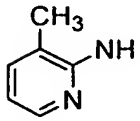
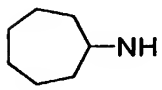
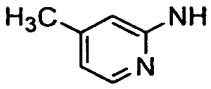
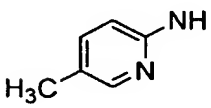
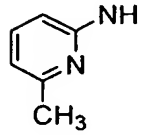
A general aspect of the invention relates to the use of compounds of a bisphosphonic acid derivative for the inhibition of growth or delaying of growth of calcium pyrophosphate, in vivo or in vitro.

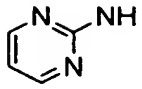
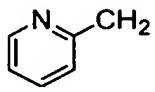
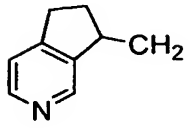
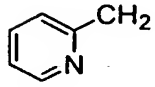
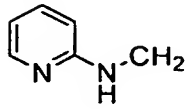
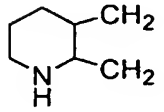
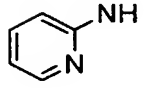
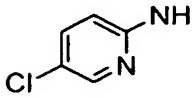
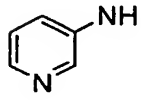
In one suitable embodiment of compounds of formula I, one or both of R^1 and R^2 is a halogen, such as a chloro group. In a further suitable embodiment one or both of R^1 and R^2 is a hydroxy group. In a further suitable embodiment one or both of R^1 and R^2 is an optionally substituted amino alkyl group. In a combination of suitable embodiments, one of R^1 and R^2 is hydroxy and the other and R^2 is a substituted alkylamino group, such as in ibandronic acid (1-hydroxy-3[methyl(pentyl)aminopropyliden]1,1-bisphosphonic acid).

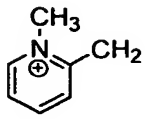
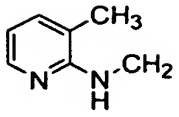
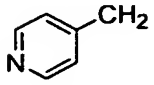
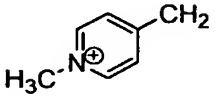
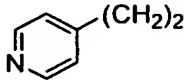
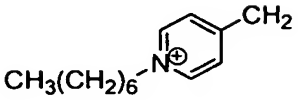
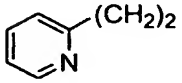
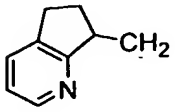
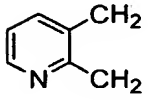
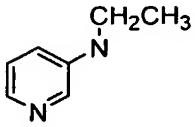
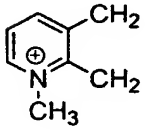
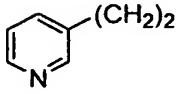
Suitable embodiments of bisphosphonates approved by the Food and Drug Administration, U.S. Department of Health and Human Services are, e.g. Didronel (etidronate), Aredia (pamidronate), Fosamax (alendronate), Skelid (tiludronate), Actonel (risedronate), Zometa (zoledronic acid), Bonefos (clodronic acid), (Bondronate) ibandronic acid, neridronate, olpadronate, incadronate, 1-Hydroxy-3-(1-pyrrolidinyl)propylidene]bisphoshonate, or [1-Hydroxy-2-imidazo-(1,2a)pyridin-3-ylethylidene]bisphosphonate. These compounds are, according to the present invention, useful for the treatment of CPDD or, as discussed infra, for the prevention or treatment of secondary caries or the treatment of a mammal with a primary carie.

In one interesting embodiment of the invention the medicament comprises a compound of formula I in combination with etidronate, pamidronate, alendronate, tiludronate, risedronate, or zoledronic acid, clodronic acid or ibandronic acid, neridronate, olpadronate, incadronate, 1-Hydroxy-3-(1-pyrrolidinyl)propylidene]bisphoshonate, or [1-Hydroxy-2-imidazo-(1,2a)pyridin-3-ylethylidene]bisphosphonate or salts thereof.

The bisphosphonates of the present invention may further be selected such that R^1 and R^2 are as outlined

R^1	R^2
	OH
$CH_3(CH_2)_4N(CH_3)(CH_2)_2$	OH
	OH
	OH
	OH
$NH_2(CH_2)_3$	OH
	H
	H
	H
	H
	H

R^1	R^2
	H
	OH
	OH
	H
	H
	(R^1, R^2)
	H
	H
	H

R^1	R^2	R^1	R^2
	OH		H
	OH	CH3	OH
	OH		OH
	OH	$NH_2(CH_2)_3$	H
$NH_2(CH_2)_2$	OH		H
	H		(R^1, R^2)
	H		
	(R^1, R^2)		
	OH		

One further interesting aspect of the invention relates to the use of Didronel (etidronate) for the preparation of a medicament for the treatment of CPDD. A further embodiment of the invention relates to the use of Aredia (pamidronate) for the preparation of a medicament for the treatment of CPDD. A still further embodiment of the invention relates to the use of Fosamax (alendronate) for the preparation of a medicament for the treatment of CPDD. A yet still further embodiment of the invention relates to the use of Skelid (tiludronate) for the preparation of a medicament for the treatment of CPDD. A yet still further embodiment of the invention relates to the use of Actonel (risedronate) for the preparation of a medicament for the treatment of CPDD. Similarly, a yet still further

embodiment of the invention relates to the use of Zometa (zoledronic acid or salts thereof) for the preparation of a medicament for the treatment of CPDD. Moreover, a yet still further embodiment of the invention relates to the use of Bonefos (clodronic acid or salts thereof) for the preparation of a medicament for the treatment of CPDD. Moreover, a yet still further embodiment of the invention relates to the use of Bondronate (ibandronic acid or salts thereof) for the preparation of a medicament for the treatment of CPDD. A yet still further embodiment of the invention relates to the use of neridronate for the preparation of a medicament for the treatment of CPDD. A yet still further embodiment of the invention relates to the use of olpadronate for the preparation of a medicament for the treatment of CPDD. A yet still further embodiment of the invention relates to the use of incadronate for the preparation of a medicament for the treatment of CPDD. A yet still further embodiment of the invention relates to the use of 1-Hydroxy-3-(1-pyrrolidinyl)propylidene]bisphoshonate for the preparation of a medicament for the treatment of CPDD. A yet still further embodiment of the invention relates to the use of or [1-Hydroxy-2-imidazo-(1,2a)pyridin-3-ylethylidene]bisphosphonate for the preparation of a medicament for the treatment of CPDD.

Among the organophosphonates further encompassed by formula I are methane-hydroxybisphosphonic acid and ethane-1-amino-1,1-bisphosphonic acid. An even more preferred organophosphonate according to the present invention is ethane-1-hydroxy-1,1-bisphosphonic acid, with the formula $CH_3C(OH)(PO_3H_2)_2$ (abbreviated EHDP). Although any pharmaceutically acceptable salt of ethane-1-hydroxy-1,1-bisphosphonic acid can be used in the practice of the present invention, the trisodium hydrogen salt, the disodium hydrogen salt, the monosodium hydrogen salt, and mixtures thereof are preferred, e.g.:

$$[CH_3C(OH)(PO_3)_2H]^{3-}, 3Na^+ \qquad (II)$$

The cation selected in salts of compounds of formula I may be any cation known to the person skilled in the art, such as mineral salts such as sodium, potassium, calcium, ammonium, typically sodium, or organic salts. The cation selected in salts of compounds of formula I may be selected for allowing or improving the solubility of the compound of formula I, particularly in vivo or for improving the release of the compound of formula I for the medicament or disintegration of the medicament.

Other pharmaceutically acceptable salts are, e.g., those described in Remington's - The Science and Practice of Pharmacy, 20th Ed. Alfonso R.Gennaro (Ed.), Lippincott, Williams & Wilkins; ISBN: 0683306472, 2000, and in Encyclopaedia of Pharmaceutical Technology.

In general, salts of phosphonic acids, such as those described herein, may crystallise as solvates, especially hydrates, which are sometimes preferred forms of the solid phosphonic acid salts due to increased stability. The compounds of the present invention, as defined by formulas I, II and elsewhere herein, may form different solvates, such as hydrates, depending on the conditions of manufacture. The invention is intended to encompass all such solvates, including for instance mono-, di-, tri-, tetra-, penta-, and hexahydrates, as well as hydrates of other stoichiometries, such as hemihydrates, and the like.

The CPDD may be due to any morphology of any form of calcium pyrophosphate such as columnar crystals, needle crystals (acicular) morphologies. Moreover, the calcium pyrophosphate may be of any hydration such as the mono-, di-, tri-, or tetrahydrate. The calcium pyrophosphate may be selected from the group consisting of triclinic and monoclinic. Preferred forms of the triclinical morphology is the dihydrate, most preferably in the form of the needle or columnar crystals. Preferred forms of the monoclinic morphology is the dihydrate or tetrahydrate. With regards to the tetrahydrate, the β- form is most preferred.

The medicament may be formulated according to conventional pharmaceutical practice, see, e.g., Remington's - The Science and Practice of Pharmacy, 20th Ed. Alfonso R.Gennaro (Ed.), Lippincott, Williams & Wilkins; ISBN: 0683306472, 2000, and in Encyclopaedia of Pharmaceutical Technology. Typically, the compounds defined herein are formulated with (at least) a pharmaceutically acceptable carrier or excipient. Pharmaceutically acceptable carriers or excipients are those known by the person skilled in the art.

As used herein, the term "pharmaceutically acceptable carrier" denotes a solid or a liquid filler or an encapsulating substance. Such substances may be selected from the group consisting of sugars such as lactose, glucose and sucrose; starches such as corn starch and potato starch; cellulose and its derivatives, such as sodium carboxymethylcellulose, ethylcellulose, cellulose acetate; gelatine, talc, malt, stearic acid, vegetable oils, polyols such as propylene glycol, polyethylene glycol, agar as well as other non-toxic compatible substances used in pharmaceutical compositions.

The administration route of the compounds as defined herein may be any suitable route which leads to a concentration in the blood or tissue corresponding to a therapeutic concentration. Thus, e.g., the following administration routes may be applicable although the invention is not limited thereto: the oral route, the parenteral route, the buccal route, the intravenous route, the cutaneous route or the nasal route. It should be clear to a person skilled in the art that the choice of administration route depends on the physico-

chemical properties of the compound together with the age and weight of the patient and on the particular the condition and the severity of the same.

The bisphosphonic acid derivatives as defined herein, or a pharmaceutically acceptable salt or hydrate thereof may be contained in any appropriate amount in a pharmaceutical composition, the pharmaceutical composition comprising an amount of about 1-95% by weight of the total weight of the composition. The composition may be presented in a dosage form which is suitable for the oral route, the parenteral route, the cutaneous route or the nasal route. Thus, the composition may be in form of, e.g., tablets, capsules, pills, powders, granulates, suspensions, emulsions, solutions, gels including hydrogels, pastes, creams, plasters, drenches, delivery devices, injectables, implants, sprays, aerosols and in other suitable form. Typically, the pharmaceutical composition comprising said compound is formulated for oral administration.

According to the present invention, for the prevention or treatment of calcium pyrophosphate deposition disease, a medically relevant concentration of the bisphosphonic acid derivative is 0.1 to 35 μM when dissolved in the animal body, typically 5 to 35 μM. Other medical relevant concentrations may however be used, depending on the individual condition of the subject, i.e. the severity and course of the disease, the subjects health and response to the particular treatment. Hence, other medical relevant concentrations of the bisphosphonic acid derivative may be lower, such as 0.5-40 μM, or higher, such as 36-50 μM.

In a suitable embodiment of the invention, the medicament comprises the bisphosphonic acid derivative in an amount so as to achieve a concentration 0.1 to 200 μM in the body, such as 0.1 to 100 μM, such as 0.1 to 50 μM, typically 0.1 to 40 μM, more typically 1 to 30 μM, most typically 5 to 30 μM.

Given the treatment may be performed in the consumption of a single unit daily dose or in multiple doses, the medicament of the invention may vary between high dosage and low dosage forms. In a suitable embodiment of the invention, the medicament comprises 5-2000 mg of the comprises the bisphosphonic acid derivative, such as 10-1000 mg, such as 20- 500 mg, such as 50-500, such as 50, 100, 150, 200, 250, 300, 350, 400, 450 or 500 mg of the compound of formula I. In a particularly suitable embodiment, the medicament comprises 50-250 mg of the comprises the bisphosphonic acid derivative.

Further embodiments of the invention relate to the use of a bisphosphonic acid derivative for the treatment of CPDD in hyaline cartilage; for the treatment of CPDD in the

fibrocartilage in the meniscus of the knee; for the treatment of CPDD in the annulus fibrosus of the intervertebral disc; for the treatment of CPDD in synovial fluid; for the treatment of CPDD in the synovium and tendon insertions; and wherein the CPDD is in the articular cartilage.

In one aspect of the present invention, the CPDD is confined to the hyaline cartilage, the fibrocartilage in the meniscus of the knee, the annulus fibrosus of the intervertebral disc, or the synovium and tendon insertions.

In another aspect of the present invention, the CPDD is especially confined to the synovial fluid or to the articular cartilage of the mammal, the mammal of which preferably is a human.

In one embodiment of the invention, the mammal to be treated has in one already suffering from deposits of calcium phosphate, and diagnosed as such. The mammal to be treated may have existing deposits, such as diagnosed as having CPDD. Thus, one interesting embodiment relates to the use of a compound of formula for the treatment of someone already diagnosed as having CPDD.

In a further embodiment, the treatment may be to a mammal not presently suffering but deemed prone to suffer from CPDD, such as to a mammal taking a medicament which has as side effects an increased likelihood of CPDD; or to a mammal over 40 years old, such as over 50, such as over 60 years old; or to a mammal whose family has a history of CPDD thus having a genetic predisposition for CPDD; or to a mammal whose lifestyle increases the likelihood of having CPDD, such as due to dietary habits or professional occupation.

Thus, a further embodiment of the invention relates to the use of a bisphosphonic acid derivative for the prophylactic treatment of CPDD.

The invention further relates to a method of treating or preventing secondary caries. In a related mode, the invention relates to a method of treating a mammal suffering from primary caries. A further aspect of the present invention relates to the use of a bisphosphonic acid derivative for the manufacture of a medicament for the prevention or treatment of secondary caries in an animal, preferentially a human.

The term "secondary caries", when used herein, is defined as caries that forms beneath, behind, in the vicinity of "primary caries", i.e. beneath, behind or in the vicinity of caries that has already formed and subsequently been treated according to any conventional

means known to the person skilled in the art. The term "caries" is intended to mean "cavities" and may also relate to softened tooth material.

An interesting aspect of the invention relates to a dental filling material, used in the treatment of caries, comprising a bisphosphonate derivative. The dental filling material may further comprise conventional materials used in tooth fillings, The secondary caries may be confined to the interface of the natural dental material (enamel, dentine, cementum and root material) and the filling material. The filling material is typically selected from the group consisting of amalgam and/or plastic but may be of any type used in dental care.

In the context of "secondary caries", it is anticipated that EHDP is to be applied onto a tooth subject to "primary caries". It is anticipated that the application of said compound is able to prevent or inhibit the subsequent formation of "secondary caries", i.e. the formation of caries below, behind or in the vicinity of the filled "primary caries". Hence, the skilled person will see that the application of EHDP to a tooth under treatment for primary caries, i.e. the application of EHDP prior to filling the primary caries with a filling material, is able to inhibit/minimize development of "secondary caries". Bisphosphonate may also be included as a depot in any type of sealing material (containing e.g. $Ca(OH)_2$ or glass ionomer containing fluoride ions) in order to inhibit/minimize development of "secondary caries".

Thus, a further embodiment of this aspect of the invention relates to the use of a bisphosphonic acid derivative for the treatment of mammal suffering from primary caries.

The bisphosphonic acid derivative used for the manufacture of a medicament for the prevention or treatment of secondary caries or for the treatment of mammal suffering from primary caries may be selected from a bisphosphonic acid derivative as defined supra.

Although any pharmaceutically acceptable salt of the compound of formula II may be used in the practice of the present invention, the trisodium hydrogen salt, the disodium hydrogen salt, the monosodium hydrogen salt, and mixtures thereof are preferred.

Alternatively stated, a further aspect of the invention is directed to a composition for the treatment or prevention of secondary caries comprising a bisphosphonic acid derivative of the formula I

$$R^1\text{-}C(PO_3H_2)_2\text{-}R^2 \qquad (I)$$

wherein R^1 and R^2 are as defined supra
or a pharmaceutically acceptable salt or hydrate thereof

Each and every embodiment of the aspect of the invention discussed in the context of the treatment of CPDD is also applicable to the treatment of secondary caries or the treatment of a mammal suffering from primary caries.

Thus, interesting embodiments of the invention the medicament comprises a compound of formula I in combination with etidronate, pamidronate, alendronate, tiludronate, risedronate, or zoledronic acid.

Furthermore, one interesting aspect of the invention relates to the use of Didronel (etidronate) for the preparation of a medicament for the treatment of treatment of secondary caries or the treatment of a mammal suffering from primary caries. A further embodiment of the invention relates to the use of Aredia (pamidronate) for the preparation of a medicament for the treatment of secondary caries or the treatment of a mammal suffering from primary caries. A still further embodiment of the invention relates to the use of Fosamax (alendronate) for the preparation of a medicament for the treatment of secondary caries or the treatment of a mammal suffering from primary caries. A yet still further embodiment of the invention relates to the use of Skelid (tiludronate) for the preparation of a medicament for the treatment of secondary caries or the treatment of a mammal suffering from primary caries. A yet still further embodiment of the invention relates to the use of Actonel (risedronate) for the preparation of a medicament for the treatment of secondary caries or the treatment of a mammal suffering from primary caries. Similarly, a yet still further embodiment of the invention relates to the use of Zometa (zoledronic acid) for the preparation of a medicament for the treatment of secondary caries or the treatment of a mammal suffering from primary caries. A yet still further embodiment of the invention relates to the use of for the preparation of a medicament for the treatment of secondary caries or the treatment of a mammal suffering from primary caries. A yet still further embodiment of the invention relates to the use of olpadronate for the preparation of a medicament for the treatment of secondary caries or the treatment of a mammal suffering from primary caries. A yet still further embodiment of the invention relates to the use of incadronate for the preparation of a medicament for the treatment of secondary caries or the treatment of a mammal suffering from primary caries. A yet still further embodiment of the invention relates to the use of 1-Hydroxy-3-(1-pyrrolidinyl)propylidene]bisphoshonate for the preparation of a medicament for the treatment of secondary caries or the treatment of a mammal suffering from primary caries. A yet still further embodiment of the invention relates to the use of or [1-Hydroxy-

2-imidazo-(1,2a)pyridin-3-ylethylidene]bisphosphonate for the preparation of a medicament for the treatment of secondary caries or the treatment of a mammal suffering from primary caries.

Other pharmaceutically acceptable salts are, e.g., those described in Remington's - The Science and Practice of Pharmacy, 20th Ed. Alfonso R.Gennaro (Ed.), Lippincott, Williams & Wilkins; ISBN: 0683306472, 2000, and in Encyclopaedia of Pharmaceutical Technology.

For the treatment of secondary caries or the treatment of a mammal suffering from primary caries, the composition may be typically be formulated in the form of a depot, paste (such as a tooth-paste), putty, rinse solution, mouth-wash, lozenge or gum.

According to the present invention, for the prevention or treatment of secondary caries or for the treatment of a patient suffering from a primary caries, a relevant concentration of the bisphosphonic acid derivative is up to the saturation point of the bisphosphonic acid derivative in the composition or solution, such as 0.001 to 50 M, such as 0.001 to 20 M, such as 0.001 to 10 M, such as 0.001 to 5 M, typically 0.01 to 5 M, more typically 0.01 to 1 M, such as 0.1 to 1 M.

The concentrations or amounts of bisphosphonic acid derivative in the composition depends on the nature of the composition and intended use. Solutions used as rinse by the dental practitioner before applying the filling (upon repair of a primary carie) may be of a different concentration than a depot to be applied between the tooth material and the filling material or of a dental filling material comprising the bisphosphonic acid derivative. Similarly, tooth pastes, lonzenges and gums are anticipated to have differing amounts or concentrations of the bisphosphonic acid derivative

An interesting aspect of the invention relates to a dental filling material, a depot, tooth-paste, rinse solution, mouth-wash, lozenge or gum comprising a bisphosphonic acid derivative, as defined herein.

In general, salts of phosphonic acids, such as those described by formula II, may crystallise as solvates, especially hydrates, which are sometimes preferred forms of the solid phosphonic acid salts due to increased stability. The salts of the compounds of the present invention, as defined by formula II and elsewhere herein, may form different solvates, such as hydrates, depending on the conditions of manufacture. The invention is intended to encompass all such solvates, including for instance mono-, di-, tri-, tetra-, penta-, and hexahydrates, as well as hydrates of other stoichiometries, such as hemihydrates, and the like.

The composition or medicaments of the invention are generally intended for the prevention of growth of calcium pyrophosphate depositions. The following Examples demonstrate, in a non-limiting fashion, the use of the compounds of formula I as a medicament as discussed supra.

Examples

The following examples are presented to illustrate the present invention and to assist one of ordinary skill in making and using the same. It will be appreciated that the examples are not intended in any way to otherwise limit the scope of the invention.

Stock crystals of m-CPPD, for use in the following examples, were prepared according to the method of Mandel et al. (Calcium pyrophosphate crystal deposition disease: preparation and characterisation of crystals, J. Crystal Growth 87 (1988) 453-462).

X-ray diffraction patterns showed that the prepared stock was mainly m-CPPD but with some t-CPPD, the most stable form of calcium pyrophosphate (CPP). The specific surface area of the m-CPPD stock is 1.35 m^2/g. The prepared crystals have a short needle-like shape with the longest dimension measuring about 10 μm.

Stock crystals of columnar t-CPPD, for use in the following examples, were prepared according to the method of Christoffersen et al. (Kinetics of dissolution of triclinic calcium pyrophosphate dihydrate crystals, J. Crystal Growth, 203 (1999) 234-243). The specific surface area of the columnar t-CPPD stock crystals is 0.8 m^2/g.

Stock crystals of acicular t-CPPD, for use in the following examples, were prepared according to the method of Christoffersen et al. (Kinetics and mechanism of dissolution and growth of acicular triclinic calcium pyrophosphate dihydrate), .R. Christoffersen, T. Balic-Zunic and J. Christoffersen, Crystal Growth and Design, 2002, 2, 567-571. The specific surface area of these stock crystals is 2.8 m^2/g.

Stock crystals of m-CPPTβ, for use in the following examples, were prepared according to the method of Christoffersen et al. (Growth and precipitation of a monoclinic calcium pyrophosphate tetrahydrate indicating auto-inhibition at pH 7, J. Crystal Growth, 212, 500-506). The specific surface area of the m-CPPTβ stock crystals is 6 m^2/g.

The compound EHDP, for use in the following examples, was in the form of a disodium salt and in the form of the tetra-acid.

Example I

Growth and formation of CPP with and without EHDP

The following examples serve to illustrate the effect that the presence of EHDP has on solutions that 1) are supersaturated (or unsaturated) with calcium pyrophosphate and 2) solutions that further comprise various stock crystals of calcium pyrophosphate dihydrate. The various stock crystals used in the following examples are: monoclinic CPPD (m-CPPD), columnar and acicular morphologies of triclinic CPPD (t-CPPD), and monoclinic CPPT (m-CPPTβ).

When calcium pyrophosphate is deposited (growth or precipitation), the solution becomes more acidic. Hence, in order to maintain a constant pH-value, a base must be added (KOH used herein). The amount of KOH added to solutions supersaturated with calcium pyrophosphate is therefore a measure of the extent and rate of the deposition of the calcium pyrophosphate crystals.

When calcium pyrophosphate is dissolved, the solution becomes more basic. Hence, in order to maintain a constant pH-value, an acid must be added (HNO_3 used herein). The amount of acid added to solutions supersaturated with calcium pyrophosphate is therefore a measure of the extent and rate of the dissolution of the calcium pyrophosphate crystals.

The supersaturation, S, is defined as

$$S = \frac{a}{a_S} = \left(\frac{IP}{K_S}\right)^{\frac{1}{3}}$$

where a is the mean ion activity, IP the activity product and K_S the solubility product. The index s refers to a saturated solution. In the following examples, the supersaturation S is calculated using an ion speciation programme "Ionics" and "Kielland" activity coefficients, the calculation of which is known by the person skilled in the art. The values of the solubility products used are: $pK_{s,t\text{-}CPPD} = 18.35$; $pK_{s,m\text{-}CPPD} = 17.6$; $pK_{s,m\text{-}CPPT} = 17.1$

Other symbols used below are defined:

C_X Total ionic concentration of X in solution

C_{EHDP} Total concentration of EHDP in system

m_0 Initial mass of crystals added

$S_{0,X}$ Initial supersaturation with respect to crystals of type X

V_{KOH}/V_H Volume of KOH or HCl added to keep pH constant

*EHDP from Tokyo Kasei Kogyo Co., Japan

Table 1 below serves to illustrate the growth of columnar t-CPPD, when columnar t-CPPD is added to 0.9 L solution supersaturated with an initial calcium pyrophosphate concentration of 0.075 mM, i.e. $C_{Ca,0} = 2C_{PP,0} = 0.15$ mM. During the experiment, pH is kept constant at 6.5 by titration with 2.0 mM KOH. $S_{0,t\text{-}CPPD} = 6.3$.

Experiment number	m_0/mg	C_{EHDP}/μM	Time / h	V_{KOH}/mL	C_{Ca} / mM
1	10.5	0	3	4.3	0.13
2	14.4	1	3	2.1	0.14
3	9.0	5	3	0	0.15

Table 1 Growth of columnar t-CPPD, pH = 6.5, temperature is 37.0 ± 0.1 °C.

From table 1 it is seen that the growth of columnar t-CPPD over 3 hours is reduced by about 50% if the concentration of EHDP (C_{EHDP}) is 1 μM, i.e. from 4.3 mL KOH to 2.1 mL KOH. When the concentration of EHDP (C_{EHDP}) is 5 μM, the rate of growth or precipitation of columnar t-CPPD is totally blocked, i.e. the pH is kept constant without the addition of KOH.

Table 2 below serves to illustrate the growth of acicular t-CPPD, when acicular t-CPPD is added to 0.9L solution supersaturated with an initial calcium pyrophosphate concentration of 0.075 mM, i.e. $C_{Ca,0} = 2C_{PP,0} = 0.15$ mM. During the experiment, pH is kept constant at 6.5 by titration with 2.0 mM KOH. $S_{0,t\text{-}CPPD} = 6.3$.

Experiment number	m_0/mg	C_{EHDP}/μM	Time / h	V_{KOH}/mL	C_{Ca} / mM
4	10.2	0	3.5	5.4	0.12
5	11.3	1	4	3.4	0.13
6	10.9	10	4	0.1	0.14

Table 2 Growth of acicular t-CPPD, pH = 6.5, temperature is 37.0 ± 0.1 °C.

From table 2 it is seen that the growth of acicular t-CPPD is inhibited by about 40% when the concentration of EHDP (C_{EHDP}) is 1 μM, i.e. from 5.4 mL KOH to 3.4 mL KOH. When the concentration of EHDP (C_{EHDP}) is 10 μM, the rate of growth or precipitation of acicular t-CPPD is substantially blocked, i.e. the pH is kept constant by the addition of 0.1 mL KOH.

Table 3 below serves to illustrate the deposition of calcium pyrophosphate (growth of m-CPPD and/or spontaneous precipitation of calcium pyrophosphate, CPP), when m-CPPD is added to 0.9 L solution supersaturated with an initial calcium pyrophosphate concentration of 0.085 mM, i.e. $C_{Ca,0} = 2C_{PP,0} = 0.17$ mM. During the experiment, pH is kept constant at 7.0 by titration with 2.0 mM KOH. $S_{0,m\text{-}CPPD} = 5.6$.

Experiment number	m_0/mg	C_{EHDP} / μM	Time / h	V_{KOH} / mL	C_{Ca} / mM
7	14.5	0	3	13.0	0.09
8	10.4	1	3	5.3	0.12
9	9.2	10	3	0	0.14

Table 3 Growth of m-CPPD, pH = 7.0, temperature is 37.0 ± 0.1 °C.

From table 3 it is seen that the growth of m-CPPD and/or precipitation of CPP over 3 hours is reduced by approximately 50% if the concentration of EHDP (C_{EHDP}) is 1 μM, i.e. from 13.0 mL KOH to 5.3 mL KOH. When the concentration of EHDP (C_{EHDP}) is 10 μM, the rate of growth or precipitation of m-CPPD is totally blocked, i.e. the pH is kept constant without the addition of KOH. Spontaneous precipitation of calcium pyrophosphate was observed after 3 h in experiment 7 without the addition of EHDP.

Table 4 below serves to illustrate the deposition of CPP when m-CPPTβ is added to 0.9 L solution supersaturated with an initial calcium pyrophosphate concentration of 0.0625 mM, i.e. $C_{Ca,0} = 2\ C_{PP,0} = 0.125$ mM. During the experiment, pH is kept constant at 7.0 by titration with 2.0 mM KOH. $S_{0,m\text{-}CPPT} = 3.1$. *EHDP from Kasei Kogyo Co., Japan is used.

Experiment number	m_0/mg	C_{EHDP}/μM	Time / h	V_{KOH} / mL	C_{Ca} / mM
10	9.8	0	4	11.5	0.053
11	11.6	1	4	10	0.062
12	9.9	5	4	0	0.11
13	10.2	10	4	0.6	0.12
14	10.9	10*	4	0.1	0.13
15	0	0	4	9	0.064
16	0	10	4	0	0.13

Table 4 Deposition of CPP (growth of m-CPPTβ and formation of CPP), pH = 7.0, temperature is 37.0 ± 0.1 °C.

From table 4 it is seen that for solutions supersaturated with calcium pyrophosphate, and with m-CPPTβ crystals added, the addition of 5 – 10 μM EHDP is able to adequately inhibit the deposition of calcium pyrophosphate, CPP, (growth of m-CPPTβ and/or the spontaneous precipitation of CPP). For example, the amount of KOH added in experiment number 10 (C_{EHDP} = 0 μM), is 11.5 ml, whereas the amount of KOH added in experiment number 12 (C_{EHDP} = 5 μM), is 0 ml, i.e., the growth of m-CPPTβ and/or the spontaneous precipitation of CPP, is severely inhibited by the addition of 5 μM EHDP.

Experiments number 15 and 16 serve to illustrate the effect when no crystals are added to a solution supersaturated with respect to calcium pyrophosphate, i.e. $C_{Ca,0} = 2C_{PP,0}$ = 0.125 mM. It is seen that the addition of 10 μM EHDP is able to inhibit fully the spontaneous precipitation of calcium pyrophosphate, i.e. the amount of KOH added is 0 mL.

Example II

Formation of CPP with and without EHDP

Tables 5 and 6 below serve to illustrate the spontaneous precipitation of calcium pyrophosphate, CPP, at room temperature, with and without the addition of 10 μM EHDP. The results from table 5 are obtained at a pH-value of approximately 7, whereas the corresponding results in table 6 are obtained at somewhat higher pH values, i.e. pH ranging from 7.0 - 7.8. pH was not kept constant in these experiments (tables 5 and 6) and no seed crystals were added.

The initial calcium concentration, $C_{Ca,0}$, is 0.9 mM in all the experiments. C_{PP} is the calcium pyrophosphate concentration, and C_{EHDP} is the concentration of the compound ethane-1-hydroxy-1,1-bisphosphonic acid. t_p is the time, after mixing EHDP and m-CPPTβ, at which the onset of precipitation is observed, i.e. as a clear decrease in the pH-value. The pH at

time t_p is pH_p. $S_{p,m\text{-}CPPT}$ is the supersaturation of m-CPPT at time t_p. When used herein, a "clear decrease" is defined as a decrease in pH of 0.5-0.6 units over a relatively short time.

Experiment number	C_{PP}/mM	$S_{p,m\text{-}CPPT}$	C_{EHDP} / µM	pH_p	t_p / h
17	0.09	6.7	0	6.7	1.3
18	0.09	6.4	10	6.7	9
19	0.18	8.6	0	6.8	1.4
20	0.18	8.2	10	6.8	5.7
21	0.36	11.2	0	7.0	1.8
22	0.36	10.6	10	6.9	4.0

Table 5 Spontaneous precipitation of calcium pyrophosphate, $C_{Ca,0}$ = 0.9 mM in all experiments number 17-22, pH ~ 7, with and without the addition of 10 µM EHDP, open to air, room temperature

From table 5 it is seen that the induction time, t_p, at which precipitation occurs from a solution supersaturated with calcium pyrophosphate, is prolonged significantly if EHDP is added. For example, the induction time is increased from 1.3 hours to 9 hours when EHDP is added in a concentration of 10 µM (compare experiment number 17 and 18).

Experiment number	C_{PP}/mM	$S_{p,m\text{-}CPPT}$	C_{EHDP} / µM	pH_p	t_p / h
23	0.09	10	0	7.4	3
24	0.09	8	10	7.0	19
25	0.18	12	0	7.5	2.8
26	0.18	10	10	7.0	22
27	0.36	14	0	7.6	4
28	0.36	14	10	7.4	8

Table 6 Spontaneous precipitation of calcium pyrophosphate, $C_{Ca,0}$ = 0.9 mM in all experiments numbers 23-28, at higher pH-values (as compared with table 5), due to exclusion of carbon dioxide by bubbling with nitrogen, with and without the addition of 10 µM EHDP, room temperature.

From table 6 it is seen that the induction times, t_p, are longer at the higher pH-values (both with and without the addition of 10 µM EHDP), although the supersaturation values

are higher. However, it is noted that the induction time for the onset of spontaneous precipitation is significantly increased upon the addition of 10 μM EHDP.

Table 7 below serves to illustrate the spontaneous precipitation of calcium pyrophosphate, CPP, from solutions supersaturated with respect to CPP, but without the addition of crystals. In the present example, pH is kept constant at pH 7.0 by the titration with 2.0 mM KOH. S_0 is the supersaturation with respect to m-CPPTβ and C_{PP} is the pyrophosphate concentration, $C_{Ca,0} = 2\ C_{PP,0}$. The induction time, t_p, denotes the time, after mixing EHDP and the solution supersaturated with CPP, at which the onset of precipitation is observed, i.e. as a clear decrease in the pH-value.

Experiment number	C_{PP}/mM	$S_{0,m\text{-}CPPT}$	C_{EHDP} / μM	t_p / h
29	0.085	3.9	0	0.5
30	0.085	3.7	10	8
31	0.17	6.2	0	8
32	0.17	6.1	10	2-5 d
33	0.32	9.3	0	7
34	0.32	9	10	15

Table 7 Spontaneous precipitation of solutions supersaturated with respect to calcium pyrophosphate, CPP, with and without the addition of 10 μM EHDP, pH = 7.0, carbon dioxide excluded, temperature is 37.0 ± 0.1 °C.

From table 7 it is seen that the induction time, at which spontaneous precipitation occurs, t_p, is longer when 10 μM EHDP is added to the solution. The largest effect is found for C_{PP} = 0.17 mM, i.e. the onset of precipitation is observed only on the course of two to five days.

Example III

Rates of dissolution of CPP with and without EHDP

Tables 8a and 8b below serve to illustrate the dissolution of acicular t-CPPD (table 8a) and columnar-CPPD (table 8b) when the mass m_0 of the respective crystal variants are added to an unsaturated solution with $C_{Ca,0} = 2C_{PP,0} = 0.07$ mM, both with and without the addition of EHDP; pH is kept constant at 5.0 by the titration of 2.0 mM HNO_3. V = 0.9 L.

Experiment number	m_0/mg	C_{EHDP} / µM	t_f / h	V_H / mL	C_{Ca} / mM
35	10.2	0	1.0	11	0.10
36	9.7	0	1.3	13	0.10
37	10	0	1.0	14.5	0.106
38	10.2	1	1.0	14.5	0.107
39	10.0	10	1.0	14.4	0.109
40	10.0	100*	1.1	13.1	0.106

Table 8a Dissolution of acicular t-CPPD, when the mass m_0 acicular t-CPPD is added to an unsaturated solution ($C_{Ca,0} = 2C_{PP,0} = 0.07$ mM), both with and without the addition of EHDP. pH is kept constant at 5.0 by the titration of 2.0 mM HNO_3. The supersaturation, S_0, is 0.50, and the temperature is 37.0 ± 0.1 °C. * EHDP from Tokyo Kasei Kogyo Co.

Experiment number	m_0/mg	C_{EHDP} / µM	t_f / h	V_H / mL	C_{Ca} / mM
41	12.3	0	1	3.3	0.08
42	10.2	10	1	2.3	0.08

Table 8b Dissolution of columnar t-CPPD, when the mass m_0 of columnar t-CPPD is added to unsaturated solutions ($C_{Ca,0} = 2C_{PP,0} = 0.07$ mM), both with and without the addition of EHDP. pH is kept constant at 5.0 by the titration of 2.0 mM HNO_3. The supersaturation, S_0, is 0.50, and the temperature is 37.0 ± 0.1 °C.

From Tables 8a and 8b it is seen that the addition of EHDP, in concentrations of up to 100 µM, has no significant effect on the dissolution rate of acicular or columnar t-CPPD.

Tables 9a, 9b and 9c below serve to illustrate the dissolution of the mass m_0 of acicular t-CPPD (Table 9a), m-CPPD (Table 9b) and m-CPPTβ (Table 9c) when added to water, pH kept constant at 7.0 by titration with 2.0 mM HNO_3. V = 0.9 L.

Experiment number	m_0/mg	C_{EHDP} / µM	t_f / h	V_H / mL	C_{Ca} / mM
43	6.5	0	2	1,4	0,005±1
44	6.9	10	3	1,4	0,003±1
45	6.8	100*	3.5	0#	0.008±1

Table 9a Dissolution of acicular t-CPPD when added to water, pH = 7.0, $S_0 = 0$. #Less acid is required because formation of soluble complexes between Ca^{2+} and EHDP release H^+. *EHDP from Tokyo Kasei Kogyo Co.

Experiment number	m_0/mg	C_{EHDP} / µM	t_f / h	V_H / mL	C_{Ca} / mM
46	12.7	0	4	4.2	0.018
47	12.8	10	4	3.3	0.016
48	12.3	100*	4	0#	0.020

Table 9b Dissolution of m-CPPD, when added to water, pH = 7.0, S_0 = 0. #Less acid is required because formation of soluble complexes between Ca^{2+} and EHDP release H^+. *EHDP from Tokyo Kasei Kogyo Co.

Experiment number	m_0/mg	C_{EHDP}/µM	t_f/h	V_H/mL	C_{Ca}/mM
49	12.1	0	4	6.8	0.028
50	11.0	10*	4	6.1	0.028
51	11.1	100*	4	≈2#	0.035

Table 9c Dissolution of m-CPPTβ, pH = 7.0, S_0 = 0. #Less acid is required because formation of soluble complexes between Ca^{2+} and EHDP release H^+. *EHDP from Tokyo Kasei Kogyo Co.

The solubilities of t-CPPD, m-CPPD and m-CPPT in water at pH = 7.0 are 0.009 mM, 0.018 mM and 0.03 mM Ca^{2+}, respectively. In Tables 9a, 9b and 9c it is seen that no significant effect of EHDP was observed, i.e. the dissolution of the added crystals is substantially the same irrespective the addition of EHDP up to C_{EHDP} = 10 mM. With C_{EHDP} = 100 mM a small increase in solubility is observed due to comlex formation.

Example IV

Use of EHDP in the context of calcium pyrophosphate deposition disease

The following example serves to illustrate the use of EHDP for the preparation of a medicament for the treatment of calcium pyrophosphate deposition disease in a mammal. It will be appreciated that the present example is not intended in any way to otherwise limit the scope of the invention.

The required dosage of the bisphosphonic acid derivative will vary with the particular condition to be treated, the severity of the of the condition, the duration of the treatment and the specific bisphosphonic acid derivative employed. However, single oral dosages of the bisphosphonic acid derivative, such as the salt of ethane-1-hydroxy-1,1-bisphosphonic acid, can range from 0.1 to 500 mg per kilogram of body weight, preferably from 0.5 to

250 mg per kilogram of body weight such as from 1.0 to 50 mg per kilogram of body weight. Said oral dosages may be administered preferably up to two times daily, such as up to three times daily, preferably such as up to four times daily. Dosages greater than, e.g., 500 mg per kilogram of body weight may produce toxic symptoms and should be avoided.

For purposes of oral administration, the active compound EHDP may be formulated in the form of capsules, tablets or granules, preferably prepared in unit dosage form together with a pharmaceutically acceptable carrier. Preferably, the pharmaceutically acceptable carrier comprises from 0.1 to 95 percent by weight of the total composition, such as from 0.1 to 98 percent by weight of the total composition.

Without being limited hereto, the active compound EHDP may also be administered parentally in aqueous solution to the subject by subcutaneous, intradermal, intramuscular or intravenous injection. Preferably, when administered parentally, the dosage may range from 0.05 to 15 mg per kilogram of body weight or such as from 0.5 to 10 mg per kilogram of body weight.

Example V

Use of EHDP in the context of secondary caries

The following example serves to illustrate the physical impact (binding strength) when adding EHDP to dental enamel imbedded in a plastic matrix, and an appropriate means for applying said compound. Hence, the example serves to illustrate the binding strength between dental enamel and a plastic filling material, when said dental enamel is treated with EHDP.

Preliminary experiments for determination of the binding strength between dental enamel treated or not treated with bisphosphonate and plastic dental filling material:

Two comparable pieces of enamel were imbedded in a plastic matrix.

1. Both pieces were polished so that a flat part of dental enamel was exposed to air.
2. Both pieces of enamel were treated with phosphoric acid, as is the normal practice when using plastic dental filling material.
3. Both pieces were rinsed under tap water.
4. One of the two pieces was treated with a few drops of 0.1 mol/L of EHDP, neutral pH, for a few seconds. This piece was then rinsed with tap water.
5. A form with a small cylindrical hole was placed on a specimen.

6. The hole was filled with a dental plastic filling material, that does not bind to the form, but which binds to the dental enamel.
7. The dental plastic filling material was polymerised by treatment with ultraviolet light, with an lamp used to polymerise this type of plastic filling in normal dental clinics.
8. After polymerisation the form was removed, leaving a small cylinder of the plastic filling material on the enamel surface of the specimen.
9. The other specimen was treated similarly.
10. Both specimens were soaked in water for some time.
11. The enamel and the plastic filling cylinder were pulled apart using an apparatus which can measure the force required to separate the two materials.
12. For both specimens the binding between enamel and plastic material was broken cleanly.
13. The force per unit area required for the specimen not treated with EHDP was 16.5 MPa. The force per unit area required for the specimen treated with EHDP was 18.7 MPa.

The result above indicates that the binding of the filling material to the tooth enamel is not weakened by the administration of EHDP to the tooth enamel. It will be appreciated that the above example could well be expanded so as to include the measurement of other parameters such as, e.g., surface roughness and adhesion. It is further anticipated that the chemical stability is able to be measured over time. Hence, it is anticipated that the chemical stability between the dental enamel and the plastic material can be monitored over time, both with and without the addition of EHDP to the dental enamel. For example, monitoring the increase or decrease of the pH in the specific environment, wherein the two components are placed, will enable the person skilled in the art to elucidate the effect of the addition of EHDP onto the dental enamel.

According to the present invention, EHDP may be applied, in an aqueous solution, onto a tooth subject to primary caries. It is anticipated, that the solution is applied onto the enamel *after* the tooth has been treated for the primary caries, according to any conventional method known to the person skilled in the art, but *prior* to the filling of the primary caries with a filling material, such as amalgam or plastic. The required dosage in the present context is such that the concentration of EHDP in the aqueous solution is in the range of from 0.001 to 5 M, preferably such as from 0.1 to 1 M. The amount administered to the caries should be in the range of from 0.5 to 5 droplets, e.g., such as from 0.0025 ml to 0.25 ml. The amount of droplets is determined by the size or extent of the caries; the requirement is that the entire surface of the caries is substantially covered by a thin layer of the aqueous solution.